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Research Paper

Histological Aspects of the Dural Crossing of Arteries to the Brain in the Fetus

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ABSTRACT

Background and Aim: This study aims to determine the histological evolution of the relationship between the dura mater and the walls of the arteries with the encephalic destination in fetuses of different ages.

Methods and Materials/Patients: We conducted a prospective descriptive study over 4 months. It included eight fetuses aged between 24 and 34 weeks of amenorrhea obtained after informed consent from the parents. Four fetuses were less than 30 weeks old and four were more than 30 weeks old. After the previously formalized anatomical dissection of the fetal skull base, partial pairs of the internal carotid and vertebral arteries were removed at the level of the dural crossing. Samples were fixed with 10% formalin and embedded in paraffin before routine histochemical (hematoxylin-eosin) and immunohistochemical (matrix metalloproteinase 7 [MMP7]) staining.

Results: We observed a "separation zone" between the dura mater and the arterial wall that is complete in fetuses between 24 and 29 weeks gestation. This cleavage zone disappeared in fetuses of 30 weeks gestation and more. This dural-arterial contact, observed from this age onwards, bears witness to a progressive arterial "invasion" by the dura mater, to reach the aspect observed in adults, characterized by the penetration of dural fibers into the arterial wall.

Conclusion: The duro-arterial relationship at the intersection level is a dynamic phenomenon that has been going on since the antenatal period.

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Highlights

• he histology of the dural crossing of the encephalic arteries was performed in the fetus. Before 30 weeks of gestational age, a zone of separation is found between the dura mater and the arterial wall.

• From 30 weeks of gestational age, adhesion is effective.

Plain Language Summary

The brain is vascularised by a pair of internal carotid arteries and another pair of vertebral arteries. These join in the cranial cavity to form the basilar artery. Initially extracranial, these arteries enter the cranial cavity after crossing the dura mater, one of the envelopes surrounding the brain. Histological studies of this crossing zone in adults showed the penetration of dural fibers into the walls of both internal carotid and vertebral arteries. Studies conducted on fetuses showed contradictory results. Some had a similar appearance to that of the adult, while others found a clear separation between the two structures.

To clarify this relationship between the dura mater and the arterial walls in the fetus, we performed a histological study on 16 dural traverses, divided into 8 pairs of internal carotid arteries and as many pairs of vertebral arteries, taken from fetuses between 24 and 34 weeks gestation. We found that in the 4 fetuses under 30 weeks, a clear separation zone was observed between the dura mater and the arterial wall, whereas after 30 weeks the dura mater adhered to the arterial wall.

Therefore, this is a dynamic phenomenon that takes place during intrauterine life. The reported differences were mostly due to differences in the age ranges of the fetuses studied.

1. Introduction

The arterial supply to the brain is provided by two arterial pairs, the internal carotid arteries and the vertebral arteries, the latter joining to form the basilar artery. Initially extra-cranial, these arteries penetrate the cranial cavity to irrigate the brain. For this purpose, they cross the meningeal envelopes, which surround this brain at the level of the foramen magnum for the vertebral arteries and the roof of the cavernous sinus for the internal carotids. It is especially the dura mater that is the most external and resistant. These regions are the site of multiple pathologies, including tumors and malformations [1, 2]. If in adults, the strong adhesion of the dura mater to the walls of these arteries and the penetration of dural fibers exists, histological studies have shown that the dural fibers penetrate the wall of these arteries at the level of these crossings, the histological aspect of this crossing in the fetus is controversial. Some have found the aspect of the adult, while others have noted a separation between these two structures [3-8].

This study was conducted to describe the histology of this crossing zone in fetuses of different ages.

2. Materials and Methods

This was a descriptive, prospective study. It was conducted from June to September 2021 (four months), at the Brazzaville University Hospital Center (Laboratory of Anatomy and Pathological Cytology) and the Faculty of Health Sciences of the Marien Ngouabi University (Laboratory of Anatomy and Organogenesis, Laboratory of Histology-Embryology).

It concerned eight heads of fetal specimens, formalized, not injected, taken from fetal cadavers after the informed consent of the parents. Eight pairs of distal dural rings were collected for the carotid artery (dura mater between segments C3-C4) [1, 9] and eight pairs of dural portions between segments V3-V4 of the vertebral arteries [10, 11] which is a total of 32 samples.

The specimens were taken from the anatomy and organogenesis laboratory of the Faculty of Health Sciences, first in the form of osteoduro arterial blocks obtained from skull bases after removal of the brain, then slow and meticulous dissection of the above-mentioned

structures. They were placed in vials containing 10% formalin. They were then processed in the Anatomy and Pathological Cytology Laboratory according to the usual cutup technique (embedding in paraffin blocks, sectioning the blocks with a microtome, placing them on slides, and staining). Haematoxylin-eosin was used to stain the slides in histochemistry. This stained the dura mater very pale orange-pink and the muscle cells a bright pink.

Slides from two fetuses were selected from the histochemical staining for the immunohistochemical technique. This was carried out in the histology-embryology laboratory from specimens that were kept at a temperature below 4°C for a maximum of 3 days. Matrix metalloproteinase 7 (MMP7), a nuclear marker of common connective tissue, was used for this process.

The slides were read using a LEICA DM 1000 LED microscope, equipped with a LEICA EC3 camera with X10, X20, and X40 magnifications.

3. Results

This study recruited 5 male fetuses and 3 female fetuses, whose sex ratio was 1.6. The mean gestational age was 29.62 weeks ranging from 24 to 34 weeks. The mean weight of the anatomical subjects was 1315 g with a maximum between 520 and 2000 g. Microscopic examination of the histological sections of the dural crossing of these arteries allowed us to notice the following for the two arteries, both on the histochemical and immunohistochemical specimens:

In fetuses aged less than 30 weeks (Figure 1A and B), the arterial wall, a connective tissue formed by its three concentric layers (intima, media, and adventitia) is surrounded by the dura mater (another connective tissue). No contact existed between the dura mater and the adventitia (the outermost layer of the arterial wall), leaving a gap between the two in the form of a cleft.

In fetuses over 30 weeks of gestational age (Figure 2A1, A2, B1 and B2), this duro-arterial separation gap disappears in favor of duro-arterial adhesion. On the other hand, spike-like penetration of dural fibers, as observed in adults, has not been individualized. Since hematoxylin-eosin stains all fibers pink, it was difficult to specify the type of relationship between the dura mater and arterial wall.

4. Discussion

In this study, two situations were observed. The first situation was a situation observed in fetuses less than 30 weeks of gestational age where a duro-arterial separation cleft existed. This separation had been reported by Tobenas-Dujardin et al. [3] in their work on fetuses of the same age as ours. Based on this observation, they situated the penetration of dural fibers into the arterial wall, as observed in adults [4-8], in the postnatal period.

The second situation was related to fetuses over 30 weeks of gestational age, whereby this dural-arterial separation gap disappeared. The dura mater is adhered to the vascular wall. However, due to the lack of specificity of the stains used in both histochemistry and immunohistochemistry, it was not possible to confirm the penetration of dural fibers into the arterial walls, as reported in adults. Broalet E [5] reported this penetration of dural fibers in 30-40 week fetuses and adults, using only histochemistry with haematoxylin-eosin-safran as a stain. In light of our work, it appears that the studies of Tobenas Dujardin and Broalet were not contradictory but complementary and testified to the dynamic character of the duro-arterial relationship in these regions.

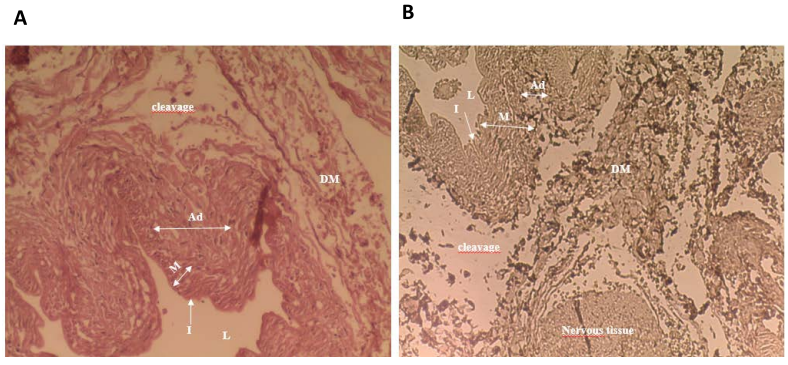

Figure 1. Histological duro-arterial section (vertebral artery) of a fetus of 25 weeks gestational age (GA)

A) HEx10, B) MMP7x10

Abbreviations: L: Lumen; M: Media; DM: Dura mater; Ad: Adventitia.

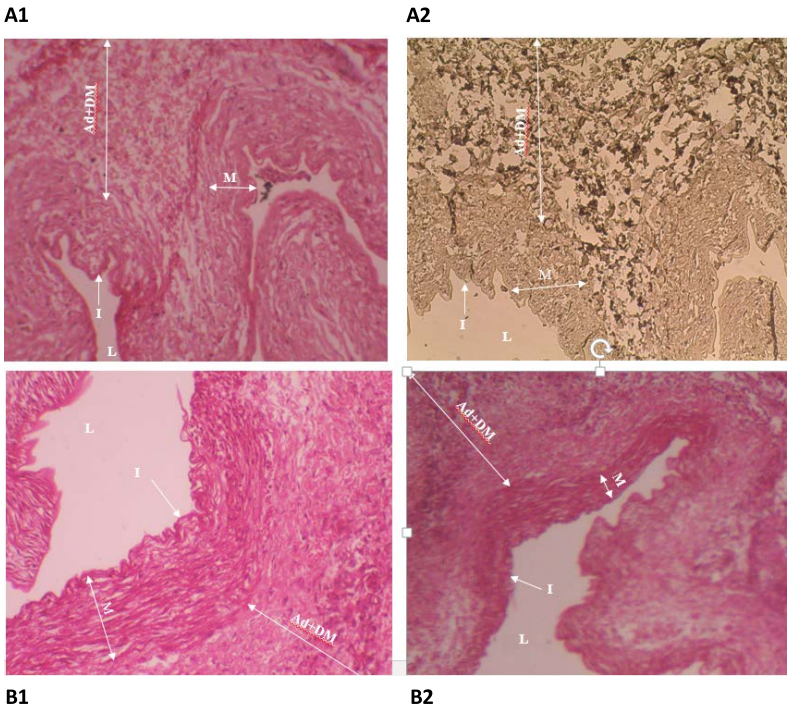

Figure 2: Duro-arterial histological section on a fetus of 32 weeks GA

A) Vertebral artery: A1) HE x10, A2) MMP7 x10)

B) Internal carotid artery: B1) HEx20, B2) HEx10)

Abbreviations: L: lumen; M: Media; DM: Dura mater; Ad: Adventitia

According to the study conducted by O'Rahilly R and Müller F [12], this complementarity is even more evident. The latter reported that the process of dural formation began on the lateral aspect of the prosencephalon and continued progressively toward its base. Therefore, it approaches the arterial walls of the brain. The separation cleft seen in fetuses less than 30 weeks is the stage before the penetration of dural fibers into the arterial wall seen in fetuses over 30 weeks.

The role of the intimate relationship between the dura mater and the arteries of the brain is not well known [7]. Hypotheses regarding this role have been proposed, arterial nicking and protection, regulation of intracranial haemodynamics, or simple phylogenetic inheritance.

5. Conclusion

The histological study of the dural crossing of arteries destined for the brain in the fetus places the period of adhesion of dural fibers to the arterial walls from the age of 30 weeks. However, the role of this entanglement of dural fibers in the vascular walls is still at the hypothesis stage.

Ethical Considerations

Compliance with ethical guidelines

During its ordinary session on December 8, 2021 and in accordance with its internal regulations, the Ethics Committee for Research in Health Sciences (cerssa), Faculty of Health Sciences, Marien Ngouabi University, Brazzaville, Republic of Congo examined the thesis entitled "histological aspects of the dural crossing of arteries destined for the brain". After deliberation, the Ethics Committee granted ethical clearance.

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Authors' contributions

Conceptualization and study design: Leon Boukassa; Data collection: Ruth Ibara Wame and Rell Boukaka Kala; Data analysis and interpretation: Fabien Gael Mouamba; Drafting the article: Leon Boukassa and Ruth Ibara Wame; Critically revising the article: Hugues Brieux Ekouele-Mbaki, Gedeon Colin Thouassa; Re-

viewing submitted version of manuscript: Sinclair Brice Kinata Bambino and Olivier Brice Ngackosso; Approving the final version of the manuscript: Leon Boukassa and Hugues Brieux Ekouele-Mbaki.

Conflict of interest

The authors declared no conflict of interest.

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